REMARKS

Entry of this supplementary amendment and reconsideration of the above-identified application in view of the amendments above and the remarks following is respectfully requested.

This document is prepared in light of an Advisory Action mailed on July 3, 2006. In said action, Examiner has advised that the use of the term "bait" may be potentially used as an art-defined term to distinct the claimed invention over the art of Takamaru and Moon. The Examiner has further indicated that the art rejection under Takamaru and Moon can alternatively be overcome by filing of a Declaration under 37 CFR 1.131 to potentially establish prior invention. The Examiner has further indicated that the rejections under 35 USC 112 second and first paragraph should overcome by the present amendments.

Claims 1, 2, 8, 9, 10, 11 and 17 have now been amended.

35 U.S.C. § 102(a) Rejections

The Examiner has rejected claims 1, 2, 6-8, 27-29 and 33-35 under 35 U.S.C. § 102(a) as being anticipated by Takemaru and Moon, The Journal of Cell Biology 149(2), April 17, 2000. The Examiner's rejections are respectfully traversed. Claims 1 and 9 have now been amended.

The Examiner points out that Takemaru and Moon teach a method of identifying interactions between polypeptides comprising use of cdc25-2 yeast strain. The cells were transfected with a first polynucleotide under control of an inducible promoter encoding a polypeptide capable of interacting with a plasmalemma. The construct is library cDNA fused to v-Src myristoylation sequences and under control of the galactose inducible promoter. The cells were transfected with a second polynucleotide comprising a fusion of a second polynucleotide and a Ras cytoplasmic mutant. Cells were grown under inductive conditions, minimal galactose. The Examiner points out that the difference between the two points indicates an interaction between the first and second polypeptide.

The ability to identify protein interactions of membrane proteins is limited by prior art approaches. This may be explained by the fact that expression of these proteins in the yeast nucleus renders these proteins non-functional due to improper

folding. The RRS system invented by the present inventor and practiced by Takamaru and Moon is based on the translocation of a cytoplasmic Ras to the plasma membrane via protein-protein interaction. Ras membrane recruitment results in activation of a viability pathway in yeast. However, the RRS system is limited in its ability to use membrane proteins as bait. This is due to the fact that fusion of a "bait" membrane protein to Ras will result in its membrane translocation independent of protein-protein interactions. Hence, the RRS system and specifically, the system described by Takamaru and Moon can only address interactions of soluble proteins and not membrane anchored proteins. Indeed, in the system described by Takamaru and Moon the "bait" (known) protein is beta-catenin which is a soluble cell signaling protein. It should be stressed that should Takamaru and Moon have used a membrane anchored protein as bait, Ras activity would have been restored independently of protein-protein interactions. Thus, the RRS system shown by Takamaru and Moon addresses an entirely different set of proteins than the claimed invention, essentially cytoplasmic proteins.

In sharp contrast to the RRS system (practiced by Takamaru and Moon) and in order to overcome its limitations, the present inventors devised a novel revolutionary approach which is specifically designed for the use of membrane proteins (polypeptides capable of interacting with the plasmalemma of the cell) as bait (known protein). In this system the known membrane protein is not fused to Ras to prevent Ras activation in the absence of protein-protein interactions. This approach is specifically designed for membrane receptors, ion channels and transporters which span the membrane even several times. Expression of these proteins in their natural environment preserves their unique three dimensional binding surfaces and allows the identification of true physiological interactions.

Indeed using the claimed method, the present inventors were able to identify a number of novel interactions with the membrane-coupled protein Chp, which were unknown at the time of filing of the present invention.

In order to better define the claimed invention over the prior art, Applicant has elected to amend claims 1, 2, 9-11 and 17 to include the limitation that the first polypeptide capable of interacting with the plasmalemma of the cell is the known

bait protein and the second polypeptide is the prey protein, emphasizing that the claimed invention is directed at the identification of membrane protein interactions.

Support for this claim amendment can be found in page 17 line 16 and page 6 lines 2-6 of the instant application.

Notwithstanding the above, Applicant wishes to draw Examiner's attention to the fact that Takamaru and Moon did not use a double inducible promoter system, resulting in a high level of false-positive interactions. Thus, Takemaru and Moon do not describe or suggest using double inducible promoter system (see new claims 50-51 and amended claims 18, 27) which enables the distinction between cells exhibiting Ras activity which results from expression and thus interaction with the prey polypeptide and a Ras activity which results from interaction-independent mobilization of Ras to the plasmalemma (i.e., false positive).

The following exemplifies the significance of using double inducible promoters. Empiric results suggest that about 5 % of the cDNAs fused to defective Ras will result in translocation of Ras to the plasma membrane. This is due to the fact that the cDNA encodes either a membrane protein or a protein that associates with the membrane or other membrane protein. Therefore, the fusion to defective Ras will result in Ras membrane translocation independent of protein-protein interaction.

Following a library screen, the clones encoding such cDNA (5% of the library) will be isolated and their growth test will always be galactose dependent. Following a library screen of a million transformants, 50,000 (!) clones will be isolated due to this false positive event. The analysis of this enormous number of clones is technically laborious and time consuming.

Applicant has now elected to amend claims 18 and 27 to using such a double-promoter selection for better distinguishing the present invention from the prior art.

Thus, the presently claimed invention addresses this shortcoming, by incorporating a second inducible promoter that will express the bait protein as well under different selection (a second inducible promoter).

Support for this claim amendment can be found in Page 27, lines 3-5 of the instant application.

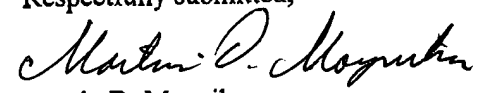
Thus, the present invention addresses interactions between a completely different set of proteins (membrane proteins) than that taught by the prior art (intracellular proteins) while obtaining a significantly lower level of false interactions due to the use of a double promoter system.

While strongly traversing the above reference, Applicant, in order to simplify the issues, attaches herewith a Declaration under 37 C.F.R. §1.131 by Applicant Ami Aronheim in which he shows a reduction to practice of the claimed invention prior to the effective publication date of Takemaru and Moon, which is the April 17, 2000. Briefly (see page 4 of the correspondence with the Editor of Nature Biotechnology) described is a novel approach to study protein-protein interactions for membrane proteins. The bait (known) protein is expressed in its natural environment, the membrane, while the prey (second polypeptide) is fused to the cytoplasmic mutant Ras. Protein interaction results in Ras membrane recruitment and activation of viability pathway. The dual inducible expression system and its significance are also described.

In view of the above declaration, the reference to Takemaru and Moon is not prior art reference relative to the instant application, and the sole rejection of the claims over Takemaru and Moon is no longer valid and should be withdrawn.

In view of the above amendments and remarks it is respectfully submitted that claims 1-2, 6-11, 15-20, 24-29 and 50-51 are now in condition for allowance. Entry of the foregoing amendment and prompt notice of allowance is respectfully and earnestly solicited.

Respectfully submitted,



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August 14, 2006
Encl.
Declaration under 37 C.F.R. §1.131 signed by Ami Aronheim
Email Document of Ami Aronheim